

Preliminary investigations of Spirulina effect on cancer cells: interest for long-term manned space missions

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Background. In view of long haul space exploration missions, the development of regenerative life support systems is of crucial importance to increase the crew autonomy and decrease the cost associated to the mass embarked. Therefore, in the late 80's, the European Space Agency initiated the MELiSSA project (Micro-Ecological Life Support System Alternative). MELiSSA has been conceived as a micro-organisms and higher plant process enabling high recycling efficiency. The cyanobacteria *Arthrospira sp.* is occupying one of the MELiSSA compartments. Its genome is now being sequenced and this will help to better understand or improve its food value as well as to have a look at its putative toxic potential. **Aim.** In this study, we were interested in studying the threshold of intrinsic cytotoxic effects of Spirulina (dry extract from Sigma containing washed and lyophilized mixed *Arthrospira* strains) on human cancer cells and its cell type dependency. **Method.** For that purpose, we used flow cytometry to estimate cell death (apoptosis and necrosis) in three human leukaemic cell lines (HELA : cervix carcinoma; IM-9 : multiple myeloma; K562: chronic myelogenous leukaemia). Cells were cultured in the presence of an aqueous extract of Spirulina (concentrations ranging from 0 to 500 $\mu\text{g/ml}$) for 15 to 40 hours. Apoptosis and necrosis were evaluated by annexin-V-PI staining, cell size and granularity. Early apoptosis was monitored by analysing the maintenance of mitochondrial membrane potential (DioC₆(3)) and the production of superoxides (hydroethidine), whereas advanced apoptosis was studied by measuring the propensity of the sub-G1 peak, esterase activity (fluorescein diacetate) and intracellular pH (carboxyfluorescein diacetate). Intracellular calcium concentration, intracellular thiols and H₂O₂ concentrations, Bcl-2 production and caspase-3 activity were also monitored. **Result.** The three cell lines had different sensitivities to Spirulina extracts : the HELA cells were most resistant, more than the K562 cells, and the IM-9 cells were most sensitive. **Conclusion.** These results suggest that, depending on the concentration, Spirulina powder extracts could enhance apoptosis in cancerous cell lines. Current work is under way to address the intrinsic toxicity of Spirulina on normal cells. Further work on cancer and normal cells will also be needed on the MELiSSA *Arthrospira sp.* PCC8005 strain. In this context, genomics should help to see which determinants are involved, and if some of them are implied in the synthesis of antioxidants able to target at cancerous

cells.